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RHINOLOGY

# Subconscious olfactory influences of stimulant and relaxant odors on immune function

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**Abstract** Brain and immune system are linked by bidirectional pathways so that changes of the central nervous system may influence various immune functions. The olfactory system may be involved in this interaction. In most odor studies subjects are aware of an odor exposure, using frequently high odor concentrations or long-term exposures without controls. In this pilot study, the potential immune effects of short-term odor exposure were examined in 32 blinded subjects (16 male, 16 female). Subjects were exposed without their knowledge either to a stimulant essential oil (grapefruit, fennel, pepper), a no-odor control or a relaxant essential oil (lavender, patchouli, rose) during a set of psychological questionnaires for 30 min at three separate visits. Activity of neutrophil granulocytes (CXCL8 release, CD16) and peripheral blood concentrations of mainly neutrophil-related immunological markers were measured. We tested the triple of stimulant odor, control and relaxant odor for every subject in a model which assumed opposite effects of the stimulant and the relaxant odor. This hypothesis was falsified by our experimental data, as no significant effect was observed for the parameters tested. The human immune functions tested in our study are not modulated by short-term odor exposure in blinded subjects. Further studies should directly dissect possible differences between long-term and short-term exposures of non-blinded subjects versus blinded subjects.

**Keywords** Olfactory · Immunology · Subconscious · Odors · Essential oils · Neutrophils

## Introduction

Brain and immune system are linked by bidirectional pathways so that changes of the central nervous system may influence various immune functions [1–3]. The olfactory system seems to be involved in this interaction, as various animal studies point to immuno-modulatory effects of odor cues: For instance, during cohabitation with tumor-bearing mice, their odor cues have induced an increase of motor activity and turnover of hypothalamic noradrenaline in other mice, indicating a higher arousal state. A decrease of neutrophil activity has been observed, showing a significant reduction of neutrophil-induced oxidative burst as well as a decrease in both the percentage and intensity of neutrophil phagocytosis [4, 5]. Exposure of male mice with female odor stimuli has led to migration of macrophages and neutrophils to the upper airways enhancing the resistance to influenza virus infection [6]. Another study with mice has shown that long-term inhalation of e.g. lemon has counteracted parts of the stress-induced immunosuppression [7]. Rats, inhaling linalool for 2 hours to reach stress repression, had a changed gene expression of whole blood cells such as neutrophils and lymphocytes [8]. In general, essential oils such as lavender (and its main ingredient linalool), patchouli and rose are said to have the main action due to relaxation as well as to sedative actions on the nervous system, presumably under control of parasympathic nerves.

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Other essential oils such as grapefruit, fennel, pepper, estragon and rosemary may have the main action of stimulating the nervous system under sympathetic control [9–12]. Thus, independent from chemosignals between mammals and pheromones, also other odors such as essential oils may take effect on important components of the immune system such as neutrophils [8] and related immunological markers. These effects may be triggered by the autonomic nervous system.

In humans, Atsumi and Tonosaki [9] have examined odor effects on the stress-related hormone cortisol in saliva and the salivary free radical-scavenging activity, preventing oxidative stress caused by free radicals and reactive oxygen species, e.g. in inflammatory processes. Sniffing lavender and rosemary for 5 min by non-blinded subjects has resulted in an increase of free radical-scavenging activity and in a decrease of cortisol levels, thus protecting the body from oxidative stress. In another study, smelling lavender has decreased significantly chromogranin A, another salivary stress marker, but not salivary cortisol, measured after 10 min [13]. Komori and coworkers [14] have used citrus fragrance to normalize neuroendocrine hormone levels and immune function in depressive patients. However, Kiecolt-Glaser and coworkers [15] have observed no reliable change of interleukin 6 and interleukin 10 production, salivary cortisol, heart rate and blood pressure, using lavender or lemon following a stressor. Thus, human studies about odor effects on varying components of the immune system show contradictory results.

Data are very limited regarding odor effects on immunological parameters, although some essential oils are regularly used in aromatherapy with the presumption of immune consequences [16–18]. Most human studies relate essential oils to putative central nervous system or cardiovascular reactions according to their classification as sedative or stimulant odors, but do not include immunological measurements. Although placebo and expectancy mechanisms may have extensive effects on odor responses, few studies have been blinded regarding the expectancies of the study and many have assessed only a single odor without any control conditions [19]. Furthermore, subjects have not been blinded regarding the existence of an odor itself in most studies performed in the past. But the sense of smell acts regularly also on a subconscious level [20, 21], and the olfactory nerves are directly linked to various parts of the central nervous system such as the limbic system and the hypothalamus [22, 23]. Subliminal odors such as pheromones have been shown to influence physiological state, mood and behavior [24]. Thus, it is not clear if subconscious exposure to odors lead to alterations of immune functions in humans.

In this pilot study, the potential immune effects of short-term inhaled essential oils were examined in blinded subjects. Subjects were exposed without their knowledge, thus on a subconscious (subliminal) level, either to a stimulant odor, a no-odor control or a relaxant odor during a set of psychological questionnaires for 30 min at three separate visits. Peripheral blood neutrophils as crucial part of the innate immune system and neutrophil-related peripheral blood cytokines were focused in this study. As this experimental design included the comparison of immune effects of a so-called stimulant odor versus a relaxant odor in every single subject tested, this study was also intended for clarifying the assumption of opposed effects of stimulant and relaxant odors in humans.

## Materials and methods

### Ethics statement

The experiments were performed according to the Helsinki Declaration. Written informed consent was obtained from all subjects before the preliminary examination for each subject. Experiments and form of consent were approved by the local ethics committee.

### Study subjects

Thirty-two young healthy subjects (European 16 male, 16 female; median age 29 years; range 20–45 years; median body mass index 27.8 kg/m$^2$) were recruited for the study. The subjects were recruited by notice (advertisement) and blinded with respect to the aim of the study (see *Cover story*). Out of more than 200 subjects, depending on a short telephone survey, 48 were invited for preliminary examination. On this first morning, young healthy subjects were selected by using a variety of interdisciplinary medical exclusion criteria and examinations. In detail, we checked the following exclusion criteria: history of chronic inflammatory diseases, smelling or taste disorder, sinus surgery, neurologic or psychiatric illness, gastrointestinal and eating disorder and surgery (also detection of Helicobacter pylori, but not history of appendectomy), diabetes and other endocrinological disorders (e.g. polycystic ovarian syndrome), impaired nasal breathing, BMI $< 20$ kg/m$^2$, regular tobacco use, pregnancy or breastfeeding, most kinds of medication (e.g. contraceptives) and acute cold or infection. Subjects aged less than 18 or above 45 years (because of age-related smelling impairment and immunologic changes) were excluded.

All subjects underwent an otorhinolaryngological and general medical examination. Colonization with Helicobacter pylori was excluded by a $C^{13}$ urea breath test. Peripheral blood measurements included a differential hemogram, HbA1c, fasting blood sugar, creatinine, hepatic

enzymes, testosterone, sex hormone-binding globulin and thyroid-stimulating hormone. Urine was checked by test strips, especially regarding glucosuria. The subjects had to give self-ratings on 100 mm visual analog scales about various abilities including their sense of smell and nasal breathing. Health questionnaires were used to screen for possible psychological disorders. Altogether, all criteria mentioned resulted in exclusion of 16 subjects for further examination. Two female subjects stopped their participation after the first main examination day, so that their results contribute only by part to the analysis.

### Course of main examination

After the preliminary examination, included subjects were examined at three independent days with a mean inter-examination period of 35 days. Women had to come within the first week of menstrual cycle. All sessions started between 8:00 and 9:30 a.m. Subjects were requested to be in a fasting condition for a minimum of 10 h. An intravenous approach was used for taking blood samples. A first course of blood samples was taken before the subjects had spent 30 min on a set of psychological questionnaires (see *Cover story*). Therefore, subjects entered an identical looking room next-door where they were exposed to an odor during that period of time at two of their three examination days. After that, they left the room and a second course of blood samples was taken. At the end of the third examination day, subjects were asked about various details of the examination room and tests, especially to evaluate if an odor was noticed and remembered. Finally, olfactory function was assessed by using Sniffin'Sticks (Burghart Instruments, Wedel, Germany) [25] and the essential oils used in the study to exclude a relevant smelling impairment and as part of another questioning.

### Odor exposure

Every subject was exposed one day to a so-called "stimulant" (="activating") odor, one day to no odor (control) and one day to a "relaxant" (="sedative") odor. We used six essential oils in this study which we received from Symrise GmbH & Co. KG (Holzminden, Germany) including gas chromatographic analyses of all oils: "grapefruit" (grapefruit oil Florida), "fennel" (fennel oil sweet fly) and "pepper" (pepper black oil) were judged as "stimulant" odors, "lavender" (lavender oil Mont Blanc), "patchouli" (patchouli oil genuine) and "rose" (rose oil Bulgarian) were judged as "relaxant" odors. 12 subjects (6 male, 6 female) were assigned to "grapefruit" and "lavender", 12 subjects (6 male, 6 female) to "fennel" and "patchouli", and 8 subjects (4 male, 4 female) to "pepper" and "rose". Essential oils were stored in aerosol cans, released with a set of well-defined spray puffs by a machine constructed by Symrise GmbH & Co. KG and then spread into the examination room by a fan. This procedure was performed shortly before entrance of the subject. Before spreading the odor, the examination was freshly ventilated to reduce potential indoor pollution.

In experiments performed before study start, other blinded and non-blinded subjects were exposed to different concentrations of the six essential oils used. Finally, respective concentrations for this study were chosen so high that they were surely perceived by non-blinded subjects for a period of 30 min, but mostly not noticed by a blinded subject entering the examination room. Indeed, only 2 of the 32 subjects included in this study mentioned a smell at one of the three examination days, but without understanding this fact as essential part of the study. The weight of the aerosol cans was measured every day so that the following amounts of essential oil were spread into the examination room (volume 50 $m^3$) [g]: "grapefruit" 1.54 ± 0.36, "fennel" 2.11 ± 0.50, "pepper" 0.62 ± 0.16, "lavender" 3.46 ± 0.37, "patchouli" 0.51 ± 0.07 and "rose" 1.98 ± 0.10.

### Cover story

All subjects were blinded to the aim of the study. A cover story was used suggesting that the primary aim of the study was a questionnaire evaluation. At every examination day, the subjects had to deal with a fixed set of multiple-choice health questionnaires for 30 min. The time of potential odor exposure passed without need of other actions. To avoid high mental stress, subjects were instructed that they should answer rather spontaneously and that they should not expect to complete all questionnaires in the time given. At the end of the last day, each subject had to judge the questionnaires on 100 mm visual analog scales (100 = highest, 0 lowest value regarding the attribute), e.g. yielding to median 86 regarding understandability and median 10 regarding stress inducing. Afterwards, every subject was informed about the odor exposure and about the aim of the study (informed consent).

### Non-cellular peripheral blood analyses

Blood was processed at a temperature below 4°C at all time points including centrifugation and then stored at −80°C until analysis. For the following parameters, the EDTA sample tubes (Sarstedt, Nümbrecht, Germany) were prepared with protease inhibitor Pefabloc SC (Carl Roth, Karlsruhe, Germany) and Dipeptidyl peptidase 4 inhibitor (Millipore, Billerica, MA). Plasma concentrations of CXCL8 (interleukin 8), CCL3 (MIP-1$\alpha$), CCL4 (MIP-1$\beta$), CCL5 (RANTES), tumor necrosis factor alpha, plasminogen activator inhibitor 1, eotaxin and interleukin 6 were

quantified with commercially available multiplex bead-based sandwich immunoassays (Bio-Plex Chemokine and Cytokine Assay and Bio-Plex Diabetes Assay, Bio-Rad Laboratories, Hercules, CA, USA) and Bio-Plex System Reader (Bio-Rad Laboratories). The assays were performed according to the manufacturer's protocols, and data were analyzed using Bio-Plex Manager software 4.11. Serum cortisol was measured by an automated analyzer.

### Cellular peripheral blood analysis

Differential hemogram was determined with an automated analyzer (Sysmex XE5000, Sysmex Corp., Kobe, Japan). For isolation of neutrophils from peripheral blood, we used previously established protocol [26]. For flow cytometric analysis, isolated neutrophils were stained with anti-CD16 PE 1:40 (clone 3G8, BD Bioscience, Heidelberg, Germany), and anti-CD66b FITC 1:100 (clone 80H3, Beckman Coulter, Fullterton, CA, USA). Analysis was performed using FACSCalibur flow cytometer (BD Bioscience). The term "Delta Median" was used to describe the median fluorescence intensity (median fluorescence intensity of the specific antibody minus median fluorescence intensity of the isotype control). To measure constitutive or induced CXCL8 release by neutrophils, purified neutrophils ($1 \times 10^6$/mL) were cultured in the presence or absence of 10 ng/mL lipopolysaccharide (LPS) (kindly provided by Prof. Dr. K. Brandenburg, Research Center Borstel, Borstel, Germany) or 100nM N-Formyl-Met-Leu-Phe (fMLP) (Sigma-Aldrich, Taufkirchen, Germany) for 24 h in RPMI-1640 (Invitrogen, Karlsruhe, Germany) supplemented with 10% fetal calf serum (Biochrom, Berlin, Germany), and 1% Penicillin/Streptomycin (Invitrogen). CXCL8 was determined by enzyme linked immunosorbent assay (ELISA) (R&D Systems, Minneapolis, MN, USA) according to the manufacturer's protocols.

### Statistical analysis

Standard descriptive statistics was used to summarize the data (e.g., means and standard deviations). We used generalized linear mixed models (with random intercepts for each subject and fixed effects for the categorized odors). Stimulant odor, control and relaxant odor were integrated into this model with assumption of opposite effects of the stimulant and the sedative odor. The paired Student's $t$ test was used to assess between group differences (only in the Supplementary Data).

All reported $P$ values are nominal, two-sided and not adjusted for the testing of multiple hypotheses. We applied a significance level $\alpha$ of 0.05 for each statistical test. SPSS Version 16 (SPSS Inc., Chicago, IL, USA) was used for statistical analyses.

## Results

### Composition of essential oils

Each essential oil used in the study was analyzed by gas chromatography. The main components of each essential oil and their percental contents are given in Table 1. Terpenes and sesquiterpenes such as limonene, pinene, myrcene, carvone, guajene, caryophyllene and carene, ethers such as anethole, alcohols such as citronellol, nerol, geraniol, patchoulialcohol and linalool were detected. In a typical manner, the essential oils consisted of a variety of these single components with separate properties given in the literature regarding their odor and their potential physiological and psychological effects [10, 12, 17]. The components of the essential oils were similar to former studies [11], but differed in their percentages. Thus, potentially similar effects of the essential oils used in this study could be assumed and compared with studies in the past.

### No significant immunological effect in a combined model of all odors

Blinded subjects were exposed without their knowledge either to a stimulant essential oil (grapefruit, fennel, pepper), a no-odor control or a relaxant essential oil (lavender, patchouli, rose) during a set of psychological questionnaires for 30 min at three separate visits. Peripheral blood concentrations of various immunological markers and activity of neutrophils (CXCL8 release, CD16) were measured. The triple of stimulant odor, control and relaxant odor for every subject was combined in a statistical model with assumption of opposite effects of the stimulant and the relaxant odor. The results of this statistical analysis are given in Table 2. In the model used, odor exposure did not yield to a significant effect for any parameter measured. Plasminogen activator inhibitor 1 may have the tendency to be increased by stimulant and decreased by relaxant essential oils ($P = 0.061$). In the subgroup of female subjects, potential effects of essential oils may be assumed for plasminogen activator inhibitor 1 ($P = 0.028$) and tumor necrosis factor $\alpha$ ($P = 0.037$). We performed an analysis of every single odor to screen for single essential oils and parameters potentially useful in further studies. A selection of results is shown in the Supplementary Data, but does not claim to include satisfactory statistic evidence.

Altogether, the analysis of that combined model, using all odors together, is not consistent with the hypothesis of opposed effects of stimulant versus relaxant odors on neutrophil-related cytokines and neutrophil activity in the peripheral blood of blinded subjects.

**Table 1** Gas chromatographic analyses of essential oils

| Essential oil | Components | Contents (%) | Essential oil | Components | Contents (%) |
|---|---|---|---|---|---|
| Grapefruit | Limonene | 90.0 | Lavender | Linalool | 38.0 |
| (A1) | Menthadiene | 1.1 | (S1) | Linalylacetate | 34.0 |
| | Myrcene | 0.9 | | Borneol/Lavandulol | 3.4 |
| | Carvone | 0.7 | | *β-cis*-Ocimene | 2.8 |
| | Limonenepoxide | 0.6 | | Caryophyllene | 2.6 |
| | *trans*-Carveol | 0.6 | | Lavandulylacetate | 2.0 |
| | α-Pinene | 0.4 | | *β-trans*-Ocimene | 1.8 |
| | Menthadiene | 0.4 | | Eucalyptol | 1.7 |
| | *cis*-Carveol | 0.3 | | Terpinenol-4 | 1.7 |
| | Sabinene | 0.3 | | Myrcene | 1.2 |
| Fennel | Anethole | 71.5 | Patchouli | Patchoulialcohol | 33.3 |
| (A2) | Fenchone | 12.4 | (S2) | δ-Guajene | 14.5 |
| | α-Pinene | 3.8 | | α-Guajene | 12.5 |
| | Limonene/β-Phellandrene | 3.5 | | Seychellene | 7.1 |
| | α-Terpineol | 3.2 | | α-Patchoulene | 5.0 |
| | α-Phellandrene | 1.8 | | Caryophyllene | 3.5 |
| | β-Pinene | 0.7 | | Aciphyllene | 3.1 |
| | Myrcene | 0.6 | | γ-Patchoulene | 2.4 |
| | Campfer | 0.3 | | Pogostol | 2.2 |
| | *para*-Cymol | 0.3 | | β-Patchoulene | 2.0 |
| Pepper | Caryophyllene | 27.2 | Rose | Citronellol/Nerol | 46.1 |
| (A3) | Limonene/β-Phellandrene | 14.8 | (S3) | Geraniol | 17.3 |
| | δ-3-Carene | 8.5 | | Nonadecane | 7.8 |
| | Sabinene | 8.0 | | Nonadecen-9 | 3.1 |
| | β-Pinene | 7.9 | | Heneicosane | 3.0 |
| | α-Pinene | 7.2 | | Heptadecane | 2.8 |
| | α-Copaene | 3.0 | | Eugenolmethylether | 1.5 |
| | δ-Elemene | 2.5 | | *trans*-Citral | 1.2 |
| | Humulene | 1.7 | | Linalool | 1.2 |
| | Myrcene | 1.5 | | *cis*-Citral | 0.9 |

Given are the ten components of the essential oil with the highest content in the gas chromatographic analysis

## Discussion

This study tested if short-term subconscious exposure to odors may lead to alterations of neutrophil-related immune functions in humans. Therefore, the potential immune effects of inhaled essential oils were examined in blinded subjects. If the triple of stimulant odor, control and relaxant odor for every tested subject was combined as a model with assumption of opposed effects of the stimulant and the relaxant odor, no significant effect was observed for all immunological parameters analyzed. Thus, short-term subconscious odor exposure may have no relevant effect on human immune functions.

As various studies point to potential effect of odors on the immune system as well as on the autonomic nervous system (see "Introduction"), the different results observed in this study may be based on the choice of odors, the consciousness regarding the odor, human animal differences, potentially wrong assumptions regarding the mediation of "odor-to-immune" effects and potentially on special conditions in this study regarding the subjects examined and the course of examination.

Essential oils contain a variety of ingredients with independent pharmacological properties and different single odors. Many factors regarding the origin and the farming of the plants as well as the extraction of essential oils lead to extent differences in the percentages of single components in the essential oil [17]. The odor of the essential oil as composition of the single odors is not only a simple sum of these single odors. In this explorative study, we have selected essential oils which have been used in other human or animal studies [7, 9, 11, 15]. Although analysis of essential oil composition is missing in many studies, the components of the essential oils in this study

**Table 2** Summary statistics for generalized linear mixed models analysis

| Marker | All subjects | | | Subgroup male | | | Subgroup female | | |
|---|---|---|---|---|---|---|---|---|---|
| | Direction | Slope | *P* value | Direction | Slope | *P* value | Direction | Slope | *P* value |
| *In vitro* | | | | | | | | | |
| CXCL8 release | | | | | | | | | |
| Unstimulated | +/− | 28.9 | >0.2 | − | −47.5 | >0.2 | +/− | 109.8 | 0.058 |
| LPS-induced | +/− | −115.4 | >0.2 | − | −608.8 | >0.2 | + | 441.4 | >0.2 |
| fMLP-ind. | +/− | 334.4 | 0.122 | +/− | 364.1 | 0.175 | +/− | 295.4 | >0.2 |
| D-Median CD16 | + | 3 | >0.2 | + | −15.1 | >0.2 | − | −34 | >0.2 |
| *Plasma/serum* | | | | | | | | | |
| CXCL8 | − | −0.4 | 0.196 | − | −0.6 | 0.191 | − | −0.2 | >0.2 |
| CCL3 | − | −0.9 | >0.2 | − | −1.3 | >0.2 | − | −0.4 | >0.2 |
| CCL4 | − | −0.6 | >0.2 | − | −0.6 | >0.2 | − | −0.7 | >0.2 |
| Cortisol | − | −12.8 | 0.132 | − | −15.1 | >0.2 | − | −10.4 | >0.2 |
| PAI-1 | + | 237.8 | 0.061 | + | 76.8 | >0.2 | + | 417.4 | 0.028 |
| TNF-alpha | + | 1 | 0.185 | − | 0 | >0.2 | + | 2.1 | 0.037 |
| IL-6 | + | 0.5 | >0.2 | +/− | 0.2 | >0.2 | + | 0.7 | 0.164 |
| Eotaxin | − | −2.2 | 0.165 | − | −1.5 | >0.2 | − | −3 | >0.2 |
| CCL5 | +/− | −2.4 | >0.2 | − | −2.7 | >0.2 | + | −1.9 | >0.2 |

The triple of stimulant odor, control and relaxant odor for every subject was combined as a model with assumption of opposed effects of the stimulant and the relaxant odor. This results in a line with a median (describing the direction of the effect), a slope (describing also the presumed strength of the effect) and the related *P* value. Direction (median): "+" means that stimulant odors increase and relaxant odors decrease the parameter given, "−" means an effect in the opposite way, and "+/−" means that statistical analysis yielded no clear direction. As example, a simplifying explanation is also given for PAI-1 in the Supplementary Data (Figure 3, Legends)

(Table 1) seem to be similar to former studies [11], but different in their percentages. The impact of these differences is not clear, but we assume potentially similar effects of the essential oils used in this study compared with studies in the past.

The effects of essential oils depend on the way of application and their concentration. High-concentrated absorption or intake of essential oils by inhalation, ingestion or epidermal application with presumably measurable blood concentrations may mediate effects rather in a pharmacological way. In opposite, smelling the odors of essential oils in low concentrations may take effect mainly through the olfactory and the central nervous system [15]. The olfactory perception and the central processing depend on characteristics of the subject such as gender [27, 28] or cultural origin [29] and on the evaluation of the odor such as its pleasantness [30–32]. For example, pleasant and unpleasant odors are linked to different brain regions [30]. Attention to the affective value alters how the brain processes olfactory stimuli [33, 34]. Importantly, the sense of smell acts regularly also on a subconscious level [20, 21]. Thus, in opposite to most studies regarding odor effects, in our study the subjects were blinded both to the aim of the study and to the existence of an odor. Furthermore, we used a short-term odor exposure of 30 min in opposite to long-term inhalation studies such as Fujiwara et al. [7] and accepted the possible effects of sensory adaptation [35]. This different study design may explain the missing observation of odor effects in our study. Also the contrary results between other studies [9, 11, 15] may be due to differences in methods.

In agreement with the literature, the statistical analysis of our study based on the assumption that there are two groups of essential oils with contrary effects regarding stimulating effects and immune-activation. If this assumption is wrong for a part of the essential oils used, the combined statistical model used may not lead to clear results. We were aware of this problem within the scope of an explorative study. We also pursued the goal to search for an essential oil that may have more extended effects than the other essential oils and hence could be used preferably in future studies. An analysis of the single odors used was performed (Supplementary Data). Some potential tendencies were observed (under reserve of the deficient group sizes for single odors), but also without clear principle regarding the attribution of the essential oil to the two groups described. However, future studies using larger sample size must be performed to exclude false-positive effects due to variability in the control group (e.g. Supplementary Figure 2B, middle panel). Furthermore, more permissive exclusion criteria, potentially the use of an immunologic stimulus, repeated or varyingly concentrated

odor exposures and different stimulation times may be useful in future studies. The potential mechanisms of a subconscious odor exposure are not equal to a conscious odor exposure with simply lower odor concentrations [24]. In the past, most odor studies were focused on a conscious odor exposure or did not consider that problem. In a future study design, it is strongly recommended that investigators either compare conscious and subconscious odor exposures or clearly decide which kind of exposure is preferred.

Altogether, on the one side, subconscious short-term odor exposure in humans may have no relevant effect on human neutrophil-related immune functions. Alternatively, the concept of opposed effects of stimulant and relaxant odors is not relevant in this relation. On the other side, this explorative study encourages further studies how conscious and unconscious odor perception is linked to the immune system as well as to the central nervous system. This possibly leads to novel insights useful for the general application of odors and flavors as well as for the general understanding of the interaction between sensory inputs, brain and immune system. Ideally, this results in the chance of modulating the immune system by the use of odors.

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